# First Year Qualifying Exam

Wei Hu 16124676

June 24, 2016

## 1 Abstract

Studies have shown that D-CARB is a potent inhibitor of intestinal and colon carcinogenesis in animal models and a mixture of D-CARB and aspirin has an interaction effect in mouse models. In our study, we have mainly two primary objectives. One is determine if D-CARB is effective in reducing polyp development and more or less effective with aspirin usage. The other one is to quantify the impact of D-CARB on hearing loss. Data is collected from 384 patients and 184 patients randomly selected from the whole. For the first objective, we build two Poisson models.Analysis and results show that D-CARB is negative associated with rate of adenomas. the rate of polyp development in the group with treatment of D-CARB is estimated to be 92% (95%CI: 78% - 97%) lower than the rate of polyp development in the group with treatment of a matched placebo where two groups have the same aspirin usage. Meanwhile, the association varies across different usage of aspirin with a p-value of 0.068. For the second objective, a linear mixed effects model is proposed. Results show that there exists effect of D-CARB on hearing loss with a significant p-value of 0.035. Among patients who are not aspirin users, the mean value of hearing detection increases by 0.40(dB) when comparing two groups , one group has treatment of D-CARB and the other group has treatment of a matched placebo.

# 2 Introduction

Cancer is a primary cause of death in the United States. Studies showed that removing of precursor adenomas is a way to prevent colorectal cancer and diet and inflammation are associated with risk of it. However, these results do not contribute a lot to medical practice by various reasons. Then scientists turn attention to D-CARB, a potent inhibitor pf intestinal and colon carcinogenesis in animal models. Researchers also showed that D-CARB and aspirin has an interesting mixture effect in vitro models. An important disadvantage of D-CARB is its potential effect on hearing loss showed in mouse model. This effect might be associated with the exposure time to D-CARB. In addition, [1] shows that long time usage of aspirin can affect hearing loss. But when the patients stop to use aspirin, their hearing ability can recover. In the dataset, the treatments and aspirin usage for each patient are recorded. To detect the effect on hearing loss caused by D-CARB, it is necessary to adjust for aspirin usage. Based on these findings, in our study, we have two main objectives: 1) determine if D-CARB is effective in reducing polyp development and more or less effective with aspirin usage. 2) quantify the impact of D-CARB on hearing loss.

# 3 Methods

## 3.1 Study Recruitment and Randomization

The first dataset comes from 364 high risk patients with a history of resected adenomas. Patients consist of 265(75.28%) males and 87(24.72%) females, who come from six sites in the United States. Among 364 patients, 294(84%) are whites, whereas the rest mainly consists of American Indian or Alaskan Native, Asian or Pacific Islander, Black, German, Indian, Hispanic, Spanish. Ages of patients vary from 41.44 and 79.33 which are broadly representative. To detect the efficiency of D-CARB, these 364 patients are randomly assigned into two groups with same sample size(182). One group received oral D-CARB

600 mg once daily and the other one received a matched placebo. Studies in rodent models indicate the efficiency of combination chemoprevention strategies compared to individual agents. So under each treatment, 69(37.91%)patients are aspirin users and 113 (62.09%) patients are not. To determine the development of adenomas, all the patients were required to receive annual sigmoidoscopies and the total number of adenomas from each patient was recorded over 36 month observation period.

184 patients in the second dataset form a randomly chosen subset of 384 patients in the first dataset. In this dataset, 96(52%) patients received oral D-CARB 600 mg once daily and 88(48%) patients received a matched placebo. [1] shows continuous usage of aspirin can cause hearing loss to some extent. Among 184 patients, there are 116(63%) aspirin users. All the patients were required to take audiology tests at three time points: 1)baseline (the time of randomization) 2) approximately 18 months after the baseline 3) approximately 36 months after the baseline. Date of the audiology tests were recorded and varied widely across patients. Audiology tests have 8 frequency types: 250 Hz, 500 Hz, 1000 Hz, 2000 Hz, 3000 Hz, 4000 Hz, 6000 Hz, 8000 Hz. At each visit, both tests for the right and left ear were recorded.

Since patients' age in the dataset are not integer and ages are distributed broadly, it is reasonable to make categorization by dividing them into four groups: (40- 50], (51,60], (61,70], (71,80]. One notable thing is that by **Table 4** in Appendix, all the characteristics group are approximately evenly distributed under treatment D-CARB and treatment Placebo. This indicates that it might be reasonable to only consider the usage of aspirin, two treatments in further analysis. **Table 5** in Appendix gives us a look on the number of adenomas observed varied by different treatments and aspirin usage. Among aspirin users, only 9(12.16%) adenomas were founded among those who received D-CARB treatment. For patients who did not use aspirin, only 1(1.67%) adenomas were founded in D-CARB group whereas 124(92.54%) adenomas found in the control group.

## 3.2 Statistical Methods

In this study, the first primary objective is a hypothesis analysis on determining whether D-CARB has a negative association(reduction) on polyp development and whether this association varies by the daily usage of aspirin. The response, the number of adenomas, indicates we are modeling count data. A natural way to estimate rate of recurrent adenomas is to build Poisson regression models. As common, the significance level is set to be 0.05 and two-sided p-value of less than 0.05 is considered statistical significant. To test whether the association between risk rate of adenomas and D-CARB treatment varies or not by usage of aspirin, we use Likehood Ratio Test as well as Wald Test. Since we are using generalized linear regression model, it is necessary to check overdispersion by Pearson residuals. If overdispersion exists, that is, the actual variance of response exceeds the specified variance, it is better to use robust variance estimator. In addition, I would use the deviance goodness of fit test for model diagnostics.

The second primary object is to quantify the potential association between treatment with D-CARB and hearing detection. Even though we know from wikipedia that "hearing sensitivity peaks around 3000 Hz"(https://en.wikipedia.org/wiki/Hearing_loss), I would include all the information contained in tests under all different frequencies. It is reasonable since the range of human hearing is from 20-20,000 Hz. To do this, I use the average value of hearing detection across all frequencies as the response. In the audio_long dataset, we have repeated measurements on each subjects. Therefore it is natural to build Linear Mixed Effects Model. Based on the analysis of thedataset, we have three main assumptions: 1)There is no correlation within specific subject. 2)There is no correlation in random effects cross subjects. 3)All the observations are independent.
The model is defined as:

$$Y_i(t) = \beta_0 + b_{0i} + (\beta_1 + \beta_{1A}A_i + \beta_{1D}D_i + \beta_{1AD}A_iD_i + b_{1i})t + \epsilon_i(t)$$

where $t \in \{0, 1, 2\}$(We denote each 18 mouth period as one time unit without loss of gener-

ality). $A_i = \begin{cases} 1 & \text{aspirin user} \\ 0 & \text{otherwise} \end{cases}$, $D_i = \begin{cases} 1 & \text{D-CARB treatment} \\ 0 & \text{otherwise} \end{cases}$. $\{\beta_0, \beta_1, \beta_{1A}, \beta_{1AD}\}$ are coefficients of fixed effect. $\{b_{0i}, b_{1i}\}$ are coefficients of random effect. Since we only have three time points, it is unnecessary to include correlation across all time points within one subject. So we assume $\underline{\epsilon_i} \sim$ iid Normal$(0, \sigma^2 I)$ and $\underline{b_i} = (b_{0i}, b_{1i}) \sim$ iid Normal$(0, \Sigma_b)$ where $\Sigma_b$ is diagonal matrix denoted as diag$(\sigma_0^2, \sigma_1^2)$

To test the significance of coefficients of fixed effect, I would use likelihood ratio test based on asymptotic normality of MLE. As for coefficients of random effect, it is supposed to test the null hypothesis: $\sigma_k^2 = 0$ versus the alternative hypothesis: $\sigma_k^2 > 0$. We want to perform likelihood ratio test. However the distribution of test statistic $-2log\frac{L_0}{L_1}$ is a mixture of two chi-square distribution under the null hypothesis. We have two ways to continue the test. One way is to use the results from Self and Liang (1987). Another way is to simulate the empirical distribution of test statistic.

# 4 Results

## 4.1 Association between D-CARB and polyp development

We have shown in Table 4 in Appendix that all the covariates are approximately evenly distributed by treatment with D-CARB and treatment without D-CARB. This is the intuition why we only include Aspirin and D-CARB in the Poisson model. To reinforce the conclusion, one straight forward way is to add categorical covariate Site, Age group, Sex, Ethnic to the former model respectively and perform likelihood ratio test. Results are shown in Table 6 in Appendix. All the P-value are not statistical significant. Therefore we only include Daily aspirin usage and Assigned Treatment in the model. By Pearson residuals, the dispersion parameter $\varphi$ is estimated to be 2.41. Therefore there exists overdispersion and we use robust variance estimator for further analysis. P-value in goodness of fit test is 0.15, which indicates the chosen model is reasonable. Furthermore,

from Table 1, the rate of polyp development in the group with treatment of D-CARB is estimated to be 92% (95%CI: 78% - 97%) lower than the rate of polyp development in the group with treatment of a matched placebo where two groups have the same aspirin usage. Based on this result, we can conclude that D-CARB is effective in reducing polyp development over 36 months.

Table 1: Association between D-CARB and Polyp Development

| Factor | Rate Ratio(95%CI) | P value |
|---|---|---|
| Daily aspirin use | | |
| 0(no) | 1(referent) | |
| 1(yes) | 1.33(0.85 - 2.16) | 0.25 |
| Assigned treatment | | |
| Placebo | 1(referent) | |
| D-CARB | 0.08(0.03 - 0.22) | $< 0.0001$ |

## 4.2 Variation of Association across Aspirin Usage

Our goal is to detect if D-CARB is more or less effective in patients that use aspirin daily. Figure 1 gives us a clear look on mean of adenomas under all different combinations of treatment and aspirin usage. It is obvious that two lines in the plot are nor parallel, which indicates that the effect of D-CARB on polyp development may vary under aspirin usage. What's more, the slope of line representing aspirin usage is obviously smaller than the slope of line representing no aspirin usage. This indicates that D-CARB is more effective in patients that use aspirin daily. To test the statistical significance of interaction between daily aspirin usage and assigned treatment, we perform a Wald Test resulting a p-value of 0.068. From Table 4, among patients who do not use aspirin daily, the rate of polyp development in the group with treatment of D-CARB is estimated to be 76% (95%CI: 58% - 95%) lower than the rate of polyp development in the group with treatment of a matched placebo. Similarly, among patients who use aspirin daily, the rate of polyp development in the group with treatment of D-CARB is estimated to be 98% (95%CI: 71% - 99%) lower than the rate of polyp development in the group with treatment of a matched placebo. It is easy to see that D-CARB with aspirin usage has

a better effect on reducing the rate of adenomas compared to D-CARB without aspirin usage. Therefore we have sufficient evidence that D-CARB is more effective in patients that use aspirin daily.

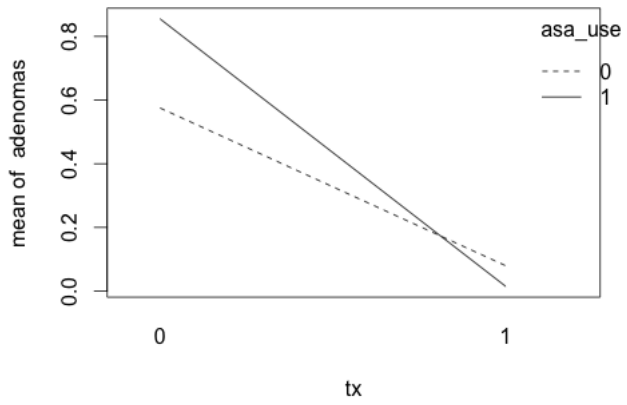


Figure 1

Table 2: Variation of Association across Aspirin Usage

| Factor | Rate Ratio(95%CI) | P value |
|---|---|---|
| Daily aspirin use | | |
| 0(no) | 1(referent) | |
| 1(yes) | 1.48(0.89 - 2.47) | 0.126 |
| Assigned treatment | | |
| Placebo | 1(referent) | |
| D-CARB | 0.14(0.05 - 0.42) | 0.0004 |
| Interaction | | |
| D-CARB:Aspirin0 | 1(referent) | |
| D-CARB:Aspirin1 | 0.122(0.012- 1.171) | 0.068 |

## 4.3 Association between D-CARB and Hearing Loss

In this part, the goal is to quantify the potential impact of treatment with D-CARB on hearing loss. [1] shows that continuous aspirin usage can affect heating loss. To detect the effect on hearing loss coming from treatment with D-CARB, it is reasonable to discuss the impact of D-CARB under the aspirin usage status respectively. Figure 2 shows the results of audiology test on 3 time points under different aspirin usage status. Figure

3 shows the average hearing detection versus different combination of aspirin usage and treatments.

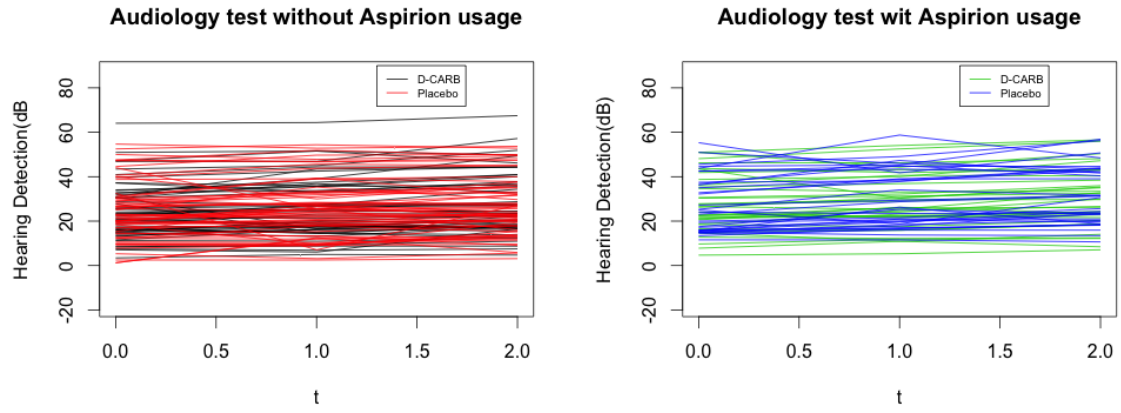


Figure 2: Plot of response with and without aspirin usage

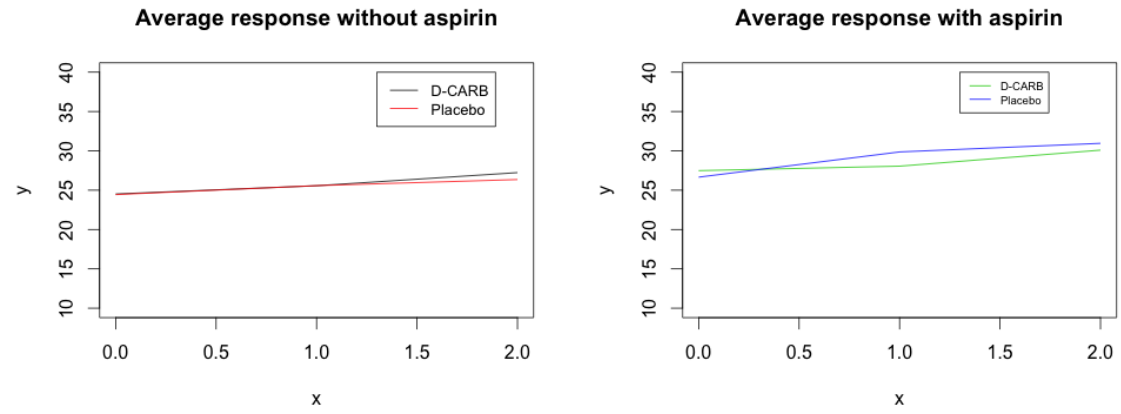


Figure 3: Plot of average response with and without aspirin usage

From Figure 2, we can find that mean trends for subjects are almost straight lines, which indicates that it is wise to choose$\{1, t\}$ as basis functions. In Figure 2, we can easily find that without aspirin usage, the average of response across all the subjects with treatment of D-CARB increases slightly faster than it without treatment of D-CARB. Meanwhile, in the other plot, the average of response across all the subjects with treatment of D-CARB increases slightly slower than it without treatment of D-CARB during the 1st time point and the 2nd time point. However after the 2nd time point, the

average of response across all the subjects with treatment of D-CARB increases slightly faster than it without treatment of D-CARB. Therefore the effect of D-CARB differs by different aspirin usage status. Based on these findings, our model is defined as bellow:

$$Y_i(t) = \beta_0 + b_{0i} + (\beta_1 + \beta_{1A}A_i + \beta_{1D}D_i + \beta_{1AD}A_iD_i + b_{1i})t + \epsilon_i(t)$$

All the assumptions are defined the same as section 4.2. Table 3 yields the estimates for the linear mixed effects model. To test if effect of D-CARB differs by different aspirin usage status, we have the null hypothesis $H_0 : \beta_4 = 0$ versus the alternative hypothesis $H_1 : \beta_4 \neq 0$. The maximum log-likelihood under $H_0$ is $-1771.988$ and the maximum log-likelihood under $H_1$ is $-1769.764$, which results a test statistic of 4.448. The test statistic has a chis-quare($df = 1$) distribution under $H_0$ and a p-value of 0.035. The p-value is significant, which reinforces the existence of interaction term in analysis before. By Figure 2, it seems no random effect in slope. To test it, we set $H_0 : \sigma_1^2 = 0$ and $H_1 : \sigma_1^2 \neq 0$. By simulation, the resulting p-value is 0.95. By results from Self and Liang (1987), under the null hypothesis, the test statistic has a distribution of $\frac{1}{2}\mathcal{X}_{(1)} + \frac{1}{2}\mathcal{X}_{(2)}$, which results a p-value of 0.804. Therefore we conclude that random effect in slope is not significant. To test there exists effect on hearing loss coming from D-CARB, we set $H_0 : \beta_4 + \beta_5 = 0, \beta_4 = 0$ and $H_1$ : at least $\beta_4 \neq 0$ or $\beta_4 + \beta_5 \neq 0$. We perform a Wald test resulting a p-value of 0.06, which is kind of significant. Therefore we conclude D-CARB has effect on hearing loss.

Regarding Table 3, among patients who are not aspirin users, the mean value of hearing detection increases by 0.40(dB) when comparing two groups , one group has treatment of D-CARB and the other group has treatment of a matched placebo. However, among patients who are aspirin users, the mean value of hearing detection decreases by 0.84(dB) when comparing two groups , one group has treatment of D-CARB and the other group has treatment of a matched placebo. It is possible that the effect of aspirin on hearing loss is so significant that explains most of the variability in response.

Table 3: Variation of Association across Aspirin Usage

| Coefficient | Estimate | Std.Error | t value |
|---|---|---|---|
| $\beta_0$ | 25.45 | 0.95 | 26.90 |
| $\beta_1$ | 0.93 | 0.28 | 3.32 |
| $\beta_2$ | 1.27 | 0.47 | 2.72 |
| $\beta_3$ | 0.40 | 0.41 | 0.98 |
| $\beta_4$ | -1.24 | 0.67 | -1.85 |

# 5 Discussion

For the first primary objective, we focus on if D-CARB has a negative association(reduction) on polyp development over 36 month and this association varies by different usage of aspirin. We obtain the conclusion that there exists a negative association and the association varies by aspirin usage. During our analysis, we exclude covariates site, patient age, gender, ethnic regarding the even distribution in two groups. Indeed, when I try to add these covariates into the model, the coefficient regarding D-CARB does not change approximately. Furthermore, due to the exclusion of unrelated covariates, it is not necessary to consider missing value anymore since missing values only appear in those covariates. The limitation is that even though it is almost evenly distributed, there is still slight difference in the distribution of covariates comparing two groups especially for age. Therefore age might be related to rate of adenomas. To test if the association varies, the p-value resulted from Wald-test is 0.068, which is significant to some extent but not significant enough. However it does not affect our final conclusion.

For the second primary objective, we pay attention to the effect of D-CARB on hearing loss. From the dataset, it only contains three time points of observations. The advantage is that we don't need to consider covariates structure(AR(1)) within one subject. However, there is an obvious disadvantage that the number of observations is too small that we don't have enough information about patients. For audiology test under different frequency, we use the average value as the response. This is reasonable since the hearing detection values under different frequency are close to each other for a specific patient.

Indeed there is another better way that can improve the accuracy. Since patients' ears are most sensitive under frequency 3000(Hz), we can use a series of kernel functions that reach the maximum on 3000. When the frequency goes far from 3000, the value of kernel function gets smaller. To choose the best kernel function, one efficient way is to use cross validation. Based on the chosen kernel function, we can assign weight to each frequency and cal calculate the weighted sum of response under all frequencies as the response in model. Results show that we must consider interaction term between aspirin usage and treatment. Even though we obtain the conclusion that D-CARB has effect on hearing loss, results also show that among patients who are aspirin users, the mean value of hearing detection decreases by 0.84(dB) when comparing two groups , one group has treatment of D-CARB and the other group has treatment of a matched placebo. The reason for this unexpected result might come from the unreasonable randomization scheme. We know that 184 patients are randomly chosen from 364 patients in the first dataset. However, the recruitment and randomization scheme are prepared for the response, the number of adenomas and the controlling group for the first primary objective. Therefore for the second primary objective, the randomization does not have any meaning.

# References


[1] McFadden D, Plattsmier HS, Pasanen EG. *Aspirin-induced hearing loss as a model of sensorineural hearing loss.*

[2] John A Baron, Susan Budinger, Nicholas Petrelli, Daniel David Karp *A Randomized Trial of Aspirin to Prevent Colorectal Adenomas in Patients with Previous Colorectal Cancer.*

# 6 Appendix

Table 4: Base-Line Characteristics of the Patients

| Characteristic | Placebo | D-CARB | Total |
|---|---|---|---|
| Sex | | | |
| Male | 134(76.14%) | 131(94.43%) | 265(75.28%) |
| Female | 42(23.86%) | 45(25.57%) | 87 (24.72%) |
| Ethnic | | | |
| American Indian or Alaskan Native | 0(0%) | 1(0.57%) | 1(0%) |
| Asian or Pacific Islander | 8 (4.55%) | 5(2.84%) | 13(4%) |
| Black | 8 (4.55%) | 6(3.41%) | 14(4%) |
| German, Indian, Hispanic, Spanish | 1(0.57%) | 0 (0%) | 1(0%) |
| Hispanic | 12(6.82%) | 13 (7.39%) | 25(7%) |
| Other | 2(1.14%) | 1(0.57%) | 3(1%) |
| Spanish | 0(0%) | 1(0.57%) | 1(0%) |
| White | 145(82.93%) | 149(84.66%) | 294(84%) |
| Site | | | |
| 1 | 34(18.68%) | 34(18.68%) | 68(19%) |
| 2 | 20(10.99%) | 20(10.99%) | 40(11%) |
| 3 | 29(15.93%) | 28(15.38%) | 57(16%) |
| 4 | 17(9.34%) | 15(8.24%) | 32(9%) |
| 5 | 25(13.74%) | 24(13.19%) | 49(13%) |
| 6 | 35(19.23%) | 37(20.33%) | 72(20%) |
| 8 | 5(2.75%) | 4(2.20%) | 9(2%) |
| 9 | 2(1.10%) | 2(1.10%) | 4(1%) |
| 11 | 15(8.24%) | 18(9.89%) | 33(9%) |
| Age group | | | |
| 41-50 | 21(11.80%) | 11(6.18%) | 32(9%) |
| 51-60 | 62(34.83%) | 66(37.08%) | 138(36%) |
| 61-70 | 69(38.76%) | 71(39.89%) | 140(39%) |
| 71-80 | 26(14.61%) | 30(16.85%) | 56(16%) |

Table 5: Comparison of Treatments

| | Placebo | D-CARB |
|---|---|---|
| Aspirin | 65(87.84%) | 9(12.16%) |
| Non-Aspirion | 59(98.33%) | 1(1.67%) |
| Total | 124(92.54%) | 10(7.46%) |

Table 6: Estimates of Covariates

| Factor | Rate Ratio(95%CI) | P value |
|---|---|---|
| Site | | |
| 1 | 1(referent) | |
| 2 | 1.65(0.74 - 3.68) | 0.22 |
| 3 | 1.52(0.70 - 3.28) | 0.29 |
| 4 | 1.06(0.40 - 2.78) | 0.91 |
| 5 | 1.31(0.58 - 2.94) | 0.52 |
| 6 | 0.89(0.38 - 2.05) | 0.78 |
| 8 | 0(0 - Inf) | 0.99 |
| 9 | 0(0 - Inf) | 0.99 |
| 11 | 0.60(0.17 - 2.08) | 0.42 |
| Age group | | |
| (40,50] | 1(referent) | |
| (50,60] | 0.79(0.35 - 1.78) | 0.56 |
| (60,70] | 0.63(0.27 - 1.45) | 0.27 |
| (70,80] | 0.60(0.22 - 1.68) | 0.33 |
| Sex | | |
| Female | 1(referent) | |
| Male | 1.07(0.55 - 2.06) | 0.85 |
| Ethnic | | |
| American Indian or Alaskan Native | 1(referent) | |
| Asian or Pacific Islander | $> 10000$(0 - Inf) | $> 0.99$ |
| Black | $> 10000$(0 - Inf) | $> 0.99$ |
| German,Indian,Hispanic, Spanish | $> 10000$(0 - Inf) | $> 0.99$ |
| Hospanic | $> 10000$(0 - Inf) | $> 0.99$ |
| Other | $> 10000$(0 - Inf) | $> 0.99$ |
| Spanish | $> 10000$(0 - Inf) | $> 0.99$ |
| White | $> 10000$(0 - Inf) | $> 0.99$ |